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FEB 27 2008

Walters et al
Ser. No. 10/537,254
Docket No. 05-084
AMENDMENT AFTER ELECTIONAmendments to the Claims

Herein, with respect to the amended claims, please note that "strikeout" matter is shown with larger-than-normal italic letters containing the strikeout horizontal marks such as in this example: *~~STRIKEOUT~~*.

Here is a listing and status of the claims provided by this AMENDMENT AFTER ELECTION.

1. (Currently amended) A method of treating vesicles with exogenous material for insertion of the exogenous material into the vesicles, comprising the steps of:

a. retaining *~~a - suspension - of -~~* the vesicles and the exogenous material in a medium in a suspension in a treatment volume in a chamber which includes electrodes, wherein the chamber has a geometric factor (cm^{-1}) defined by the quotient of the electrode gap squared (cm^2) divided by the chamber volume (cm^3),

wherein said geometric factor is less than or equal to 0.1 cm^{-1}),

wherein the suspension of the vesicles, *~~and~~* the exogenous material, and the is - in - a medium *~~which~~* is adjusted, such that the suspension medium has conductivity

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in a range spanning 0.001 to 100 ~~0 - 01 - 10 - 1 - 0~~
milliSiemens/cm,

wherein the resistance of the suspension in the chamber
is greater than one ohm,

wherein the suspension is enclosed in the chamber
during treatment, and

b. treating the suspension enclosed in the chamber with one
or more pulsed electric fields,

wherein in accordance with a. and b. above, the treatment
volume of the suspension is scalable.

2. (Original) The method of claim 1 wherein the chamber is a
closed chamber.

3. (Original) The method of claim 1 wherein the chamber has at
least a 2 milliliter capacity.

4. (Original) The method of claim 1 wherein the chamber and the
contents thereof are sterile.

5. (Original) The method of claim 1 wherein the chamber
includes entry and exit ports for entry and removal of the
suspension.

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6. (Original) The method of claim 1 wherein the electrodes are parallel plate electrodes.

7. (Cancelled)

8. (Original) The method of claim 1 wherein the electric fields include a rectangular voltage pulse waveform to produce a uniform pulse electric field between parallel plate electrodes greater than 100 volts/cm and less than 5,000 volts/cm, substantially uniform throughout the treatment volume.

9. (Cancelled)

10. (Cancelled)

11. (Cancelled)

12. (Cancelled)

13. (Cancelled)

14. (Cancelled)

15. (Cancelled)

16. (Original) The method of claim 1 wherein the pulsed electric fields are from electrical pulses which are in a sequence of at least three non-sinusoidal electrical pulses, having field strengths equal to or greater than 100 V/cm, to the material, wherein the sequence of at least three non-sinusoidal electrical

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pulses has one, two, or three of the following characteristics:

(1) at least two of the at least three pulses differ from each other in pulse amplitude; (2) at least two of the at least three pulses differ from each other in pulse width; and (3) a first pulse interval for a first set of two of the at least three pulses is different from a second pulse interval for a second set of two of the at least three pulses.

17. (Cancelled)

18. (Cancelled)

19. (Original) The method of claim 1 which is carried out in sequential batches.

20. (Original) The method of claim 1 wherein the exogenous material is a therapeutic material.

21. (Original) The method of claim 1 wherein a therapeutic product is formed from the treatment of the vesicles with exogenous material.

22. (Original) The method of claim 1 wherein the exogenous material is a polynucleotide.

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23. (Cancelled)

24. (Original) The method of claim 1 wherein the exogenous material is a polypeptide.

25. (Original) The method of claim 1 wherein the exogenous material is a protein.

26. (Cancelled)

27. (Cancelled)

28. (Original) The method of claim 1 wherein the chamber has a chamber volume, the suspension has a suspension volume, and the suspension volume is greater than the chamber volume, and wherein an initial portion of the suspension volume is moved into the chamber, retained and treated in the chamber, and moved out from the chamber, and

an additional portion of the suspension volume is moved into the chamber, retained and treated in the chamber, and moved out from the chamber.

29. (Original) The method of claim 1 wherein still further portions of the suspension volume are sequentially moved into the

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chamber, retained and treated in the chamber, and moved out from the chamber.

30. (Cancelled)

31. (Currently amended) An electroporation apparatus, comprising:

a chamber having a chamber volume of at least 2 milliliters, a pair of electroporation electrodes contained within said chamber,

an electroporation medium, carrying vesicles in suspension, contained in said chamber between said electroporation electrodes, wherein said suspension medium has a conductivity between 0.001 to 100 ~~0.01 to 100~~ milliSiemens/cm, and wherein the resistance of the suspension in said chamber is greater than one ohm.

a source of pulsed voltages electrically connected to said electroporation electrodes, and

means for adding material to said chamber for electroporation treatment therein, and means for removing treated material from said chamber.

32. (Cancelled)

33. (Cancelled)

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34. (Cancelled)

35. (Original) The apparatus of claim 31 wherein said chamber includes vent means for venting air when fluid is moved into said chamber.

36. (Cancelled)

37. (Cancelled)

38. (Original) The apparatus of claim 31 wherein said chamber includes a chamber inlet and a chamber outlet.

39. (Original) The apparatus of claim 31, further including:
a first reservoir, in fluid communication with said chamber inlet, for containing said vesicle-bearing electroporation medium prior to introduction into said chamber,

a second reservoir, in fluid communication with said chamber inlet, for containing a chamber flushing material for flushing treated vesicle-bearing medium out from said chamber, and

a third reservoir, in fluid communication with said chamber outlet, for receiving treated, vesicle-bearing medium that is flushed out from said chamber.

40. (Cancelled)

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41. (Cancelled)

42. (Previously added) The method of claim 1 wherein the time of treatment of the vesicles in the chamber is substantially the same for all vesicles.

43. (New) The method of claims 1 wherein said suspension of the vesicles, the exogenous material, and the medium is adjusted, such that said suspension has conductivity in a range spanning 0.01 to 1.0 milliSiemens/cm.

44. (New) The apparatus of claim 31 wherein said suspension has a conductivity between 0.01 to 1.0 milliSiemens/cm.